Population genetic diversity and historical dynamics of Fraser's dolphins, *Lagenodelphis hosei*

Running page head: Fraser's dolphin population genetic structure

Ing Chen[1,2], Shin Nishida[3], Lien-Siang Chou[4], Tomohiko Isobe[5,6], Antonio A. Mignucci-Giannoni[7,8] and A. Rus Hoelzel[1*]

[1] Department of Biosciences, Durham University, South Road, Durham, DH1 3LE, United Kingdom

[2] Division of Science, Yale-NUS College, 16 College Avenue West, Singapore 138527, Singapore (present address)

[3] Science Education, Faculty of Education and Culture, University of Miyazaki, 1-1 Gakuen-Kibanadai-Nishi, Miyazaki, 889-2192, Japan

[4] Institute of Ecology and Evolutionary Biology, National Taiwan University, No.1, Sec.4, Roosevelt Road, Taipei, 10617, Taiwan

[5] National Institute for Environmental Studies, 16-2 Onogawa, Tsukuba, 305-8506, Japan (present address)

[6] Center for Marine Environmental Research, Ehime University, 2-5 Bunkyo Cho, Matsuyama 790-8577, Japan.

[7] Universidad Interamericana de Puerto Rico, Centro de Conservación de Manatíes de Puerto Rico, PO Box 361715 San Juan, 00936, Puerto Rico

[8] Conservation Medicine and Ecosystem Health, Ross University School of Veterinary Medicine, PO Box 334, Basseterre, St. Kitts

[*] Corresponding author: A. Rus Hoelzel. a.r.hoelzel@dur.ac.uk

Abstract

Marine organisms face relatively few barriers to gene flow, and yet even highly mobile species such as dolphins often show population structure over regional geographic scales. Understanding the processes that promote this pattern of differentiation helps us understand the evolutionary radiation of this group, and to promote more effective measures for conservation. In this study we provide the first population genetic study of Fraser's dolphin (*Lagenodelphis hosei* Fraser, 1956), a species that was not recognized by the scientific communities until the early 1970s. We use 18 microsatellite DNA loci and one mitochondrial DNA (mtDNA) locus to compare 112 Fraser's dolphins collected in various locations, mainly from the waters off Japan, Taiwan, and the Philippines, but also including samples from the Gulf of Mexico and Caribbean Sea. Our results indicate differentiation between populations in waters off Japan, Taiwan, and the Philippines and support the findings from earlier morphological assessments for differentiation between Japanese and Philippine waters. Small sample sets also show likely differentiation between other regions in the North Pacific and North Atlantic Oceans. Moreover, the neutrality tests and mismatch analysis based on mtDNA data indicate that the populations in the western North Pacific Ocean have expanded demographically and spatially, possibly since the latest global deglaciation when sea levels and global temperatures started to rise.

Key words:

Population structure, marine mammal, Northwest Pacific Ocean, conservation, climate change

1. INTRODUCTION

Understanding population structure is essential for establishing useful inference about the process of local adaptation and evolution (Kawecki & Ebert 2004), as well as for developing conservation strategies in natural resource management (Palsbøll et al. 2007). Assessing population structure for oceanic cetaceans (whales, dolphins and porpoises) can be particularly challenging, not only because their highly dynamic open water environment usually offers little clue about potential population boundaries, but also because the population structure is often shaped in various contexts by multiple intrinsic biological factors, such as resource exploitation, physiological constraints, or behavioural/cultural stereotyping (e.g. Hoelzel 2018).

On the other hand, environmental factors such as climate change can also play a significant role in shaping marine biodiversity patterns at both regional and global scales (Renema et al. 2008, Cheung et al. 2009). Past climate oscillations have been attributed to the distribution of many contemporary cetacean species or populations, particularly for those living in middle to higher latitude waters (e.g., Hayano et al. 2004, Pastene et al. 2007, Harlin-Cognato et al. 2007, Banguera-Hinestroza et al. 2010, 2014, Taguchi et al. 2010, Amaral et al. 2012, Moura et al. 2013). However, little is reported for species from tropical waters. As modeling analyses have shown that the current global warming phenomenon could affect marine mammal diversity and distribution range globally (MacLeod 2009, Kaschner et al. 2011), further information regarding the population structure of tropical species is certainly needed.

Fraser's dolphin (*Lagenodelphis hosei*) is one of the least studied dolphin species in the world. The species was unknown to the scientific communities until

Fraser (1956) described a skull specimen collected from Sarawak, Borneo in 1895. Yet, the existence of any living Fraser's dolphins was not confirmed until the early 1970s, when further fresh specimens from the Eastern Tropical Pacific (ETP), South Africa, Australia, Taiwan and Japan, as well as sighting records of living individuals in ETP and Central North Pacific (CNP), started to emerge (Perrin et al. 1973, Tobayama et al. 1973). Further sightings, strandings and bycatch records from the North and South Atlantic Ocean were reported in the following decades (Caldwell et al. 1976, Hersh & Odell 1986, Leatherwood et al. 1993, Bones et al. 1998, Mignucci-Giannoni et al. 1999, Moreno et al. 2003, Weir et al. 2008, Gomes-Pereira et al. 2013). Fraser's dolphins are known to be widespread in pan-tropical regions of the Pacific, Atlantic and Indian Oceans, and their presence is usually associated with a particular combination of environmental characteristics: deep water environment with tropical or subtropical climate (Hammond et al. 2012, Jefferson et al. 2015, Dolar 2018). This species has been proposed to be a possible marine bio-indicator of climate change, as its recent range expansion in the North Atlantic appears to reflect the increase of regional seawater temperature in the temperate waters of the Azores (Gomes-Pereira et al. 2013).

Geographic variation for the species has been reported for pigmentation patterns (e.g. between dolphins from South Africa and the eastern Tropical Pacific; Perrin et al. 1973), body size (dolphins found off France seem to be larger than those found in the western North Pacific; Van Bree et al. 1986; however, this observation was later questioned by Amano et al. 1996), skull morphometric measurements (relatively larger and broader skills for dolphins in Japanese waters than those in Philippine waters; Perrin et al. 2003), and social assemblage (smaller pod size in the North Atlantic than in the North Pacific; Gomes-Pereira et al. 2013). However, morphological and behavioural characteristics can be plastic and may not always reflect the pattern of gene flow (West-Eberhard 1989, Crispo 2008, Prada et al. 2008).

Small sample size and sampling area coverage was also a limitation for some of these earlier studies.

Here, we assess the genetic diversity and population structure of Fraser's dolphins, with a focus on the eastern Asian regions, where this species is considered to have been negatively impacted by fisheries activities (e.g. frequent involvement in incidental or direct catches; Jefferson & Leatherwood 1994, Perrin et al. 2005, Porter & Lai 2017, Altherr & Hodgins 2018). Based on the conclusions of an earlier morphological study (Perrin et al. 2003), we hypothesized that Fraser's dolphin populations would be genetically differentiated between the Pacific and Atlantic Oceans, and between Japanese and Philippine waters. We also test the hypothesis that coincident with past periods of global warming including the last deglaciation, we may find evidence for population expansion associated with population growth in the Fraser's dolphin, consistent with that proposed for other tropical species (MacLeod 2009, Gomes-Pereira et al. 2013).

2. MATERIALS & METHODS

2.1. Sample collection, DNA fragment amplification and genotyping

The 112 samples used in this study were collected from dead Fraser's dolphins either beach-casted or perished in fishery interactions, except for three samples from Central North Pacific (CNP) which were biopsied from free-ranging dolphins (Supplementary Table S1). Based on sampling localities, we categorized the samples into seven geographic groups: Japan, Taiwan, the Philippines, CNP, ETP, Gulf of Mexico (GM), and the Caribbean Sea (CS) (Fig. 1). The species and sex identity was acquired from the archive records where the identification was based on the external morphological characters of the specimens. When in doubt, this was verified by our genetic assessments. Samples supplied by the Southwest Fisheries Science Center (USA) were titrated DNA solutions; otherwise samples were provided as a small

portion of skin or muscle tissue samples preserved in either 99% ethanol or 20% DMSO solution saturated with sodium chloride. All specimens, except the three Philippine specimens archived in es-BANK (Ehime University, Japan), were transported to and examined in the Molecular Ecology Group Laboratory at Durham University, with valid official permits issued by the authorities of Japan, Taiwan, the United States, and the United Kingdom.

The genomic DNA of tissue samples was isolated and purified using a standard proteinase-K digestion/phenol–chloroform extraction protocol (Sambrook et al. 1989). We examined 18 microsatellite loci (AAT44, D14, D22, KWM1b, KWM2b, KWM9b, TexVet5, TexVet7, MK3, MK5, Dde65, Dde69, Dde70, Dde72, Dde84, Sco11, Sco28, and Sco55; see Supplementary Table S2) and one mitochondrial DNA (mtDNA) locus (779bp of the control region using the primers described in Hoelzel et al. 1991) that have been used in earlier population genetic studies for other delphinid species, following the same procedure as described in Chen et al. (2017). Briefly, annealing was at 40^{o}C (for mtDNA) and the amplification ran for 35 cycles, with the purified product sequenced on an ABI 3730 in the forward direction. The optimal annealing temperatures and allele size ranges of each microsatellite locus are provided in Supplementary Table S2.

2.2. Microsatellite data analysis

Micro-Checker 2.2.3 (Van Oosterhout et al. 2004) was used to screen for null alleles and potential scoring errors. The R package *pegas* (Paradis 2010) was used to estimate observed heterozygosity (H_O) and expected heterozygosity (H_E), and to test for Hardy-Weinberg Equilibrium (HWE) for the sampled loci. The number of replicates for the Monte Carlo procedure was set to the default value (B = 1000). A locus was assessed for deviation from HWE using both the $\chi 2$ test and the exact test based on a Monte Carlo permutations of alleles, and excluded from further analyses if

$p < 0.001$. Inbreeding coefficient (F) was estimated for each individual using the *inbreeding* function implemented in another R package *adegenet* (Jombart 2008). Because the Japanese sample was from a single sampling event, we ran a kinship analysis using the program Kingroup (Konovalov et al. 2004) including only individuals from the Japanese sample set.

The degree of population differentiation among the geographic groups was evaluated through F statistics, and the significance was tested using G-statistic tests (Goudet et al. 1996), using functions implemented in *hierfstat* (Goudet 2005) and *pegas*, with the number of simulations set to 1000. Pairwise F_{ST} values (Nei 1987) among the three major sampling groups (i.e., the Philippines, Taiwan, and Japan) were calculated using *hierfstat*. A 95% confidential interval (CI) was generated with 1000-fold bootstrap resampling. The discriminant analysis of principal components (DAPC, Jombart et al. 2010) implemented in *adegenet* was also used to assess genetic structure and interpret individual membership. Fifteen principal components (determined according to the *a*-score analysis; Jombart et al. 2010) and 100 discriminant analysis steps were retained in the analysis. Factorial correspondence analysis (FCA) implemented in Genetix 4.0 (Belkhir et al. 2014) was applied as a complementary ordination analysis. We used the 'sur population' option since the aim was to reveal differentiation among geographic groups rather than among individuals.

Spatial population genetic structure was assessed using *Geneland* (Guillot et al. 2005). The data were analyzed using the correlated allele frequency model and the spatial model; the uncertainty associated with the spatial coordinates was set as one decimal place, the maximum rate of Poisson process was fixed to 100, and the maximum number of nuclei in the Poisson-Voronoi tessellation was fixed to 300. The number of Markov chain Monte Carlo (MCMC) iterations was set to 10^6, with a thinning at every 1000 iterations, and K was set to vary from 1 to 10. To construct the population distribution map, we set the burn-in to 200 iterations, and the spatial

domain to 174 pixels along the X-axis and 27 along the Y-axis. We also used the Mantel test implemented in *adegenet* to test the effect of isolation by distance (IBD), using both Nei's distance (non-Euclidean) and Edwards' distances (Euclidean) to estimate genetic distance, and the euclidean distance for geographic distance at the population level.

2.3. Mitochondrial DNA analysis

The mtDNA sequences were aligned and assessed using MEGA 5.05 (Tamura et al. 2011). A median-joining network was constructed using PopART (Bandelt et al. 1999, Leigh & Bryant 2015). Gene diversity (*h*), nucleotide diversity (π), Tajima's D, and Fu's Fs were estimated using DnaSP 5.10 (Librado & Rozas 2009). Historic demographic or spatial expansion was evaluated using the analysis of mismatch distributions implemented in Arlequin 3.5 (Excoffier & Lischer 2010). This was done for each putative population on its own, and for all western North Pacific samples (i.e. Japan, Taiwan and Philippines) combined as a one population. The confidence interval for the mismatch estimates was obtained from 10^4 bootstrap simulations of an instantaneous expansion under a coalescent framework. Model fit was evaluated according to the significance of the sum of square deviations (SSD) between the observed and the expected mismatch and the raggedness index (r) of the observed distribution (Harpending 1994, Schneider & Excoffier 1999).

An approximate time of expansion (*T*) was calculated through the formula $T=\tau/2u$, where τ is the simulated time of demographic or spatial expansion estimated in the mismatch analysis, and *u* is the mutation rate for the sequence in use (per locus per generation; Rogers 1995). We used an estimated generation time of 11.1 years (Taylor et al. 2007), and used two substitution rate values: 1×10^{-7} substitutions/per site/per year (Ho et al. 2011) and 7×10^{-8} substitutions/per site/per year (Harlin et al. 2003).

Arlequin was used to estimate pairwise F_{ST} and Φ_{ST}. We used the Tamura Nei model to estimate Φ_{ST} because it was the closest model available to the TVM+I model, which was suggested as the best model for our samples according to the Akaike Information Criterion (AIC) result in jModelTest 2.1.6 (Darriba et al. 2012). The level of differentiation between sample group pairs was estimated with 10^4 permutations.

3. RESULTS

3.1. Microsatellite data analysis: genetic diversity

Useful microsatellite data were obtained from 106 samples (Fig. 1; Supplementary Table S1). There were nine samples with missing data at 1—4 loci. The 18 loci examined were all polymorphic, with the number of alleles ranging from 2 to 17 (Supplementary Table S3). None of these loci showed consistent deviation from HWE across the three major sampling groups (Japan, Taiwan, and the Philippines), and so all were retained. However, for the Taiwan group, there were five loci showing signs of null alleles and deviations from HWE, though the magnitude of deviation was always small. The reason for the larger proportion of loci out of HWE in Taiwan is not known, though given that the sample size was relatively large and collected over a relatively broad temporal period (see Supplementary Table S1) a Wahlund effect is possible. Genotyping errors seemed less likely due to the overall good quality of DNA and low divergence among populations. Deviation from HWE is expected due to the Wahlund effect when differentiated populations are combined, so the higher incidence of HWE deviation for combined datasets (Supplementary Table S3) supports our interpretation of population structure (see below). Mean H_O and H_E for the three major groups ranged from 0.54 — 0.62 and 0.57 — 0.62, respectively (Table 1). The mean H_O was significantly lower than the mean H_E for the Taiwan group (upper-tailed paired *t*-test, $t = 3.58$, df = 17, p = 0.001). The Taiwan

group also showed the highest average inbreeding coefficient (F = 0.21). The kinship analysis for the Japan group showed a mean pairwise kinship of r = -0.0277, implying that within group kinship was unlikely to have affected our population-level analyses.

3.2. Microsatellite data analysis: population structure

The G-statistic test result suggested the presence of population structure in our sample (p = 0.008; Supplementary Fig. S1). Among the three groups with sufficiently large sample sizes, F_{ST} was most pronounced between the Philippines and Japan (F_{ST} = 0.013). Based on the 95% CI estimates, all pairwise F_{ST} values were significantly different from zero except the Philippines-Taiwan pair (Table 2). For regions with small sample sizes, DAPC showed that the CS samples were most distinct (Fig. 2). The three major sampling groups (Japan, Taiwan and the Philippines) could also be differentiated using both DAPC (Fig. 2) and FCA (Supplementary Fig. S2) analyses. In the DAPC group membership assignment analysis, most individuals could be reassigned to their original clusters (including all groups with small sample sizes), although some potential admixture was found among all groups including Japan, Taiwan and the Philippines (Supplementary Fig. S3).

In the Geneland analysis, K=4 was supported by the highest mean logarithm of posterior probability (LPP; Supplementary Table S4) generating a population structure pattern (Fig. 3) broadly consistent with the pattern seen in our DAPC and FCA analyses (Fig. 2, Supplementary Fig. S2). The Mantel test for isolation by distance (IBD) showed that there was no significant effect of IBD in our sample, no matter what method was used to estimate genetic distance (p = 0.948 with Nei's distance method; p = 0.897 with Edwards' distance method; Supplementary Fig. S4).

3.3. Mitochondrial DNA data analysis

We amplified a 779 bp mtDNA control region sequence in 96 samples and

identified 48 unique haplotypes characterized by 64 variable sites (Supplementary Tables S1, S5). The median-joining network showed little evidence of lineage sorting (Fig. 4). The number of haplotypes shared between Taiwan and Japan was more than that between Taiwan and the Philippines, or between the Philippines and Japan (Supplementary Table S6).

The genetic and nucleotide diversity was high for Japan, Taiwan, and the Philippines (Table 3). All three groups had a negative Tajima's D, although none of the values were statistically different from zero. With the exception of the Philippines, all Fu's Fs estimates were also negative, and the values were statistically significant in Japan and Taiwan, indicating an excess of low-frequency haplotypes, possibly resulting from an historic expansion, or selective sweep. When combining all samples from the western North Pacific together, Fu's Fs was still negative and statistically significant (Table 3).

A non-unimodal mismatch distribution was seen in Japan, Taiwan and the Philippines (Supplementary Fig. S5); however, SSD and *r* were small and statistically insignificant (Table 4), suggesting the distributions concurred with both demographic and spatial expansion models. The estimated time of population expansion was at about the same time for all three groups (Table 4), with the time of spatial expansion starting slightly later than the time of demographic expansion. The estimated chronological time for the expansion was 2,000—11,000 years ago (Table 4).

In the pairwise F_{ST} comparisons, significant differentiation was found between the Philippines and Japan ($F_{ST} = 0.033$, $p = 0.022$) and between the Philippines and Taiwan ($F_{ST} = 0.029$, $p = 0.026$) (Table 5). Comparisons among CNP, ETP, GM and CS were omitted as the sample sizes were too small to provide useful inferences. In the Φ_{ST} comparison, on the other hand, none of the paired estimates were statistically different from zero (Table 5). The exact tests based on both haplotype frequencies and the Tamura and Nei model indicated that the Philippines, Taiwan and Japan were

differentiated (Supplementary Table S7).

4. DISCUSSION

4.1. Population structure

Population differentiation between Japan and the Philippines was previously recognized from skull morphology: the skulls of Japanese samples were broader and the rostrum wider, with larger orbits and internal nares, and a longer cranium (Perrin et al. 2003). From our genetic data differentiation was evident between Japan, the Philippines (consistent with the cranial data) and Taiwan from ordination analyses with some overlap, and for F_{ST} values between Japan and the Philippines or Taiwan. This pattern was supported by the analyses in Geneland (differentiating between Japan, Taiwan, and the Philippines), but the haplotype network showed little indication of lineage sorting among any of the putative populations.

The sample size from the Philippines was comparatively small, but the pattern of differentiation detected by summary statistics (which may be affected by sample size) was generally consistent with ordination methods (which are independent of sample size with respect to the placement of individual points in Euclidean space). In general, F_{ST} values were small and of a consistent magnitude, and significantly different from zero for most comparisons among the western North Pacific putative populations. Ordination methods, which have more power, separated all groups with varying levels of overlap. For mtDNA, both the lack of lineage sorting evident in the network and the lack of significant Φ_{ST} comparisons (which reflects differences among haplotype sequences) suggest relatively recent division among populations in Japan, Taiwan and the Philippines.

A number of other marine vertebrate species inhabiting the same or adjacent regions, including common bottlenose dolphin (*Tursiops truncatus*; Chen et al. 2017), flathead mullet (*Mugil cephalus*; Shen et al. 2011) and green sea turtle (*Chelonia*

mydas; Jensen et al. 2019) also show similar patterns of structure. For the bottlenose dolphin the authors proposed that previous glacial events strengthened oceanographic barriers, with differentiation later diminished by the resumption of gene flow when the environment became favorable (Chen et al. 2017). In this study, the Philippine samples were collected from the Sulu Sea, a semi-enclosed deep-sea body of water, where most of the Fraser's dolphin sightings have been in waters 700–3500 m deep (Dolar et al. 2006, Dolar 2018). The Sulu Sea was once as shallow as 420 m or less at its edge during the glaciation epochs (Wang 1999, Voris 2000), providing the potential for habitat division during the glacial epochs.

If our samples from Japan reflect a local population, it is possible that the well documented oceanographic differences between Japanese and the waters around the Philippines or Taiwan (see Miyazawa et al. 2009) could influence dispersion and insularity. However, these samples may be from a transient or migratory population, since Fraser's dolphins are only rarely reported in the temperate waters around Japan (Amano et al. 1996, Kanaji et al. 2017). In contrast, the occurrence of Fraser's dolphins off Taiwan and the Philippines is frequently reported (Yang et al. 1999, Dolar et al. 2006, Tseng et al. 2011). The species most typically has a pan-tropical distribution in deep and offshore waters, however their more precise distributional range in the broader region is uncertain due to the scarcity of sightings in the high-seas of the western North Pacific Ocean (Kanaji et al. 2017). Therefore it is difficult to know the ranging behavior of the dolphins in our Japanese sample. Further field surveys and genetic sampling covering that region may clarify patterns of connectivity with the group of dolphins found in Japanese waters.

Limited inference for population comparisons could be drawn outside the western North Pacific Ocean as our sampling sizes were small. For instance, even though the result of our DAPC and Geneland analyses appears to support earlier morphological finding suggesting population differentiation between the Pacific and

Atlantic oceans (Perrin et al. 2003), we cannot fully exclude the possibility that this was a stochastic result due to the small number of samples (Halsey et al. 2015). Similar caution is appropriate for inference about putative population differences identified in the central North Pacific, Eastern Tropical Pacific, and Gulf of Mexico.

4.2. Population expansion history

Our mtDNA data suggest that Fraser's dolphin populations in the western North Pacific have been expanding, particularly for the population found in Japanese waters. Our estimation for the time of Fraser's dolphin population expansion in the western North Pacific is within the period of most recent deglaciation following the last glacial maximum (19,000—20,000 years ago; Clark et al. 2009), and most likely at the beginning of the Holocene (about 11,500 years ago; Mayewski et al. 2004). There is evidence for population expansions during the early Holocene for a number of cetacean species (e.g. Banguera-Hinestroza et al. 2014; Louis et al. 2014; Moura et al. 2014; Chen et al. 2017, 2018). Furthermore, there are clues suggesting range expansion for Fraser's dolphin populations in the modern age. For example, the sighting frequency of this species has increased in recent decades around the Lesser Antilles, Caribbean Sea (Watkins et al. 1994, Rinaldi & Rinaldi 2011) and the Azores (Gomes-Pereira et al. 2013). The encounter rate of stranded Fraser's dolphins in Japanese coasts has increased somewhat after the millennium (8 cases during 2000–2018 vs. 3 cases before 2000; National Museum of Nature and Science 2018). Although the trend of climate warming may be associated with these range expansions (see MacLeod 2009), it is uncertain whether the phenomenon would persist and become widespread around the globe, and what the consequences may be as this tropical species 'invades' higher latitude waters.

On the other hand, we did not detect an expansion signal for the Philippine population. The relatively high genetic diversity and flat mismatch distribution pattern

could imply a long-term stable Philippine population. However, the sampling size for the Philippine population in this study was relatively small (n = 17 for microsatellite and n = 10 for mtDNA), and the inference of population expansion was made solely based on mtDNA sequence variation. Further assessments investigating a broader range of genomic signals with more samples would reveal a more comprehensive picture for the population history of Fraser's dolphins.

4.3 Implications for conservation

Our study shows that at least for the dolphins in the western North Pacific, the mtDNA genetic diversity of the Fraser's dolphin is high compared to that of other oceanic delphinid species inhabiting the same or adjacent regions (e.g., pantropical spotted dolphin populations in Taiwan-Southern China waters: h = 0.778—0.888, π = 0.49%—0.96%, n = 4—18, Yao et al. 2004; common bottlenose dolphin populations in eastern Asian waters: h = 0.824—0.908, π = 1.368%—2.193%, n = 14—160, Chen et al. 2017). We also show that the level of diversity is similar among regions and when putative populations are pooled. High genetic diversity is consistent with large effective population size, and the potential for resilience to environmental fluctuations (Frankham 2005). However, we also find relatively fine-scale population genetic structure, and evidence for divergence among most regional population samples included in the study. This would imply a need for management strategies that protect regional diversity and the potential for local adaptation. At the same time, further systematic sampling surveys and genotyping for the dolphins in the region (especially from the Philippines) and better survey data from the Japanese region would facilitate the generation of more effective conservation management strategies.

The Fraser's dolphin is currently considered an offshore, oceanic delphinid species with least conservation concern (Hammond et al. 2012, Jefferson et al. 2015). However, the impact of frequent Fraser's dolphin bycatches (or direct catches) in the

Asian and Eastern Tropical Pacific fisheries (Jefferson & Leatherwood 1994, Perrin et al. 2005, Chou 2006, Porter & Lai 2017, Altherr & Hodgins 2018) will warrant reassessment in the context of structured populations in the western North Pacific. Given our preliminary data on differentiation among geographically distant sites, together with the data on relatively fine-scale differentiation in the western North Pacific, further samples from the Fraser's dolphin's extensive distribution range should be a priority. In particular, samples from the Eastern Tropical Pacific, South Pacific Ocean, pelagic North Atlantic Ocean and Indian Ocean, should be included in future studies to assess the species' global population structure and expansion history. If the hierarchical morphological differentiation revealed in Perrin et al. (2003) does reflect population genetic structure, then future studies should find the North Atlantic Ocean population to be the most distinctive, and possibly identify further differentiated populations in the Southern Hemisphere. We also anticipate that, by examining more Fraser's dolphin samples from a broader range, further light would be shed on the effect of global climate change on the dynamics of the world's tropical dolphin populations.

Acknowledgements

The study was funded by the International Whaling Commission Small Cetaceans Research Fund (2012/13), and the Government Scholarship for IC provided by the Ministry of Education, Government of Taiwan. The samples used in this study were provided collectively by the Cetacean Ecology Laboratory at National Taiwan University (Taiwan), es-BANK at Ehime University (Japan), and Southwest Fisheries Science Center, National Oceanic and Atmospheric Administration (USA). The authors would like to thank Kelly Robertson, Shinsuke Tanabe and the staffs at the es-BANK, Ming-Ching Lin, Wei-Cheng Yang and the students at the Aquatic Animal

Medicine Laboratory, National Chia-Yi University (Taiwan) for their assistance on sample collection, administration, and shipping. The comments offered by the contributing editor Philippe Borsa and three anonymous reviewers were much appreciated.

Availability of data and materials

The mtDNA sequences are available on GenBank (Accession numbers MN268582-MN268677). The microsatellite genotyping data will be available on request.

Authors' contributions

Designed the study: I.C., S.N. and A.R.H. Sample collection and shipping permit administration: I.C., S.N., L.-S.C, T.I. and A.A.M.-G. Statistical analyses: I.C. Wrote/revised article for intellectual content: I.C. and A.R.H. All read and approved the final manuscript.

Competing interests

The authors declare that they have no competing interests.

Ethics approval and consent to participate

The samples used in this study were collected in accordance with the regulation of local governments. Appropriate national and international permits to translocate the samples to ARH's laboratory at Durham University were obtained prior to the shipping.

Literature Cited

Altherr S, Hodgins N (2018) Small cetaceans, big problems. A global review of the impacts of hunting on small whales, dolphins and porpoises. Report of the Animal Welfare Institute, Pro Wildlife and Whale and Dolphin Conservation. 67pp. Available at https://us.whales.org/wp-content/uploads/sites/2/2018/08/small-cetaceans-big-problems.pdf (accessed on 13 March 2019)

Amano M, Miyazaki N, Yanagisawa F (1996) Life history of Fraser's dolphin, *Lagenodelphis hosei*, based on a school captured off the Pacific coast of Japan. Mar Mam Sci 12:199–214

Amaral AR, Beheregaray LB, Bilgmann K, Freitas L and others (2012) Influences of past climatic changes on historical population structure and demography of a cosmopolitan marine predator, the common dolphin (genus *Delphinus*). Mol Ecol 21:4854–4871

Bandelt HJ, Forster P, Röhl A (1999) Median-joining networks for inferring intraspecific phylogenies. Mol Biol Evol 16:37–48

Banguera-Hinestroza E, Evans PGH, Mirimin L, Reid RJ and others (2014) Phylogeography and population dynamics of the white-sided dolphin (*Lagenorhynchus acutus*) in the North Atlantic. Conserv Genet 15:789–802

Banguera-Hinestroza E, Bjorge A, Reid RJ, Jepson P, Hoelzel AR (2010) The influence of glacial epochs and habitat dependence on the diversity and phylogeography of a coastal dolphin species: *Lagenorhynchus albirostris*. Conserv Genet 11:1823–1836

Belkhir K, Borsa P, Chikhi L, Raufaste N, Bonhomme F (2004) GENETIX 4.05, logiciel sous Windows TM pour la génétique des populations. Laboratoire Génome, Populations, Interactions, CNRS UMR 5000, Université de Montpellier II, Montpellier (France). http://kimura.univ-montp2.fr/genetix/ (Accessed 9 July 2019)

Bones M, Neill B, Reid B (1998) Fraser's dolphin (*Lagenodelphis hosei*) stranded in South Ulst: first record in U.K. waters. J Zool 246:460–461

Caldwell DK, Caldwell MC, Walker RV (1976) First records of Fraser's dolphin (*Lagenodelphis hosei*) in the Atlantic and the melon-headed whale (*Peponocephala electra*) in the western Atlantic. Cetology 25:14

Chen I, Nishida S, Yang W-C, Isobe T, Tajima Y, Hoelzel AR (2017) Genetic diversity of bottlenose dolphin (*Tursiops* sp.) populations in the western North Pacific and the conservation implications. Mar Biol 164: 202

Chen I, Nishida S, Chou L-S, Tajima Y and others (2018) Concordance between genetic diversity and marine biogeography in a highly mobile marine mammal, the Risso's dolphin. J Biogeogr 45:2092–2103

Cheung WW, Lam VW, Sarmiento JL, Kearney K, Watson R, Pauly D (2009) Projecting global marine biodiversity impacts under climate change scenarios. Fish Fish 10:235–251

Chou L-S (2006) Cetacean bycatch in coastal waters of Taiwan and the ecology of Chinese white dolphins, *Sousa chinensis*. Final Report to Fishery Agency, Council of Agriculture, Republic of China (Taiwan), No. 95AS-14.1.1-FA-F2

Clark PU, Dyke AS, Shakun JD, Carlson AE and others (2009) The last glacial maximum. Science 325:710–714

Crispo E (2008) Modifying effects of phenotypic plasticity on interactions among natural selection, adaptation and gene flow. J Evol Biol 21:1460–1469

Darriba D, Taboada GL, Doallo R, Posada D (2012) jModelTest 2: more models, new heuristics and parallel computing. Nat Methods 9:772

Dolar MLL (2018) Fraser's dolphin L*agenodelphis hosei*. In: Würsig B, Thewissen JGM, Kovacs KM (eds) Encyclopedia of marine mammals, third edition. Academic Press, San Diego, CA, p 392–395

Dolar MLL, Perrin WF, Taylor BL, Kooyman GL, Alava MNR (2006) Abundance and

distributional ecology of cetaceans in the central Philippines. J Cetacean Res Manag 8:93–111

Excoffier L, Lischer HEL (2010) Arlequin suite ver 3.5: a new series of programs to perform population genetics analyses under Linux and Windows. Mol Ecol Resour. 10:564–567

Frankham, R (2005) Genetics and extinction. Biol Conserv 126: 131–140

Fraser FC (1956) A new Sarawak dolphin. Sarawak Museum J. 7(8 n.s.):478–503(pl. 22–4).

Gomes-Pereira JN, Marques R, Cruz MJ, Martins A (2013) The little-known Fraser's dolphin *Lagenodelphis hosei* in the North Atlantic: new records and a review of distribution. Mar Biodivers 43:321–332

Goudet J (2005) Hierfstat, a package for R to compute and test hierarchical F-statistics. Mol Ecol Notes 5: 184–186

Goudet J, Raymond M, de Meeüs T, Rousset F (1996) Testing differentiation in diploid populations. Genetics 144:1933–1940

Guillot G, Mortier F, Estoup A (2005) GENELAND: a computer package for landscape genetics. Mol Ecol Notes. 5:712–715

Halsey LG, Curran-Everett D, Vowler SL, Drummond GB (2015) The fickle P value generates irreproducible results. Nat Methods 12:179–185

Hammond PS, Bearzi G, Bjørge A, Forney KA and others (2012) *Lagenodelphis hosei.* In: IUCN (ed) The IUCN Red List of Threatened Species. Version 2014.3. 2012. www.iucnredlist.org. (accessed 05 Feb 2015)

Harlin AD, Markowitz T, Baker CS, Würsig B, Honeycutt RL (2003) Genetic structure, diversity, and historical demography of New Zealand's dusky dolphin (*Lagenorhynchus obscurus*). J Mammal 84:702–717

Harlin-Cognato AD, Markowitz T, Wuersig B, Honeycutt RL (2007) Multi-locus phylogeography of the dusky dolphin (*Lagenorhynchus obscurus*): passive

dispersal via the west-wind drift or response to prey species and climate change? BMC Evol Biol 7:17

Harpending HC (1994) Signature of ancient population growth in a low-resolution mitochondrial DNA mismatch distribution. Hum Biol 66:591–600

Hayano A, Yoshioka M, Tanaka M, Amano M (2004) Population differentiation in the Pacific white-sided dolphin *Lagenorhynchus obliquidens* inferred from mitochondrial DNA and microsatellite analyses. Zoolog Sci 21:989–1000

Hersh SL, Odell DK (1986) Mass stranding of Fraser's dolphin, *Lagenodelphis hosei*, in the western North Atlantic. Mar Mam Sci 2:73–76

Ho SYW, Lanfear R, Phillips MJ, Barnes I, Thomas JA, Kolokotronis SO, Shapiro B (2011) Bayesian estimation of substitution rates from ancient DNA sequences with low information content. Syst Biol 60:366–375

Hoelzel AR (2018) Molecular ecology. In: Würsig B, Thewissen JGM, Kovacs KM (eds) Encyclopedia of marine mammals, third edition. Academic Press, San Diego, CA, p 613–618

Hoelzel AR, Hancock JM, Dover GA (1991) Evolution of the cetacean mitochondrial D-loop region. Mol Biol Evol 8:475–493

Jefferson TA, Leatherwood S (1994) *Lagenodelphis hosei.* Mamm Species 470:1–5.

Jefferson TA, Webber MA, Pitman RL (2015) Marine mammals of the world: a comprehensive guide to their identification. 2nd Edition. Academic Press, San Diego, CA

Jensen MP, FitzSimmons NN, Bourjea J, Hamabata T, Reece J, Dutton PH (2019) The evolutionary history and global phylogeography of the green turtle (*Chelonia mydas*). J Biogeogr DOI: 10.1111/jbi.13483

Jombart T (2008) adegenet: a R package for the multivariate analysis of genetic markers. Bioinformatics 24:1403–1405

Jombart T, Devillard S, Balloux F (2010) Discriminant analysis of principal components: a new method for the analysis of genetically structured populations. BMC Genet 11:94

Kanaji Y, Okazaki M, Miyashita T (2017) Spatial patterns of distribution, abundance, and species diversity of small odontocetes estimated using density surface modeling with line transect sampling. Deep Sea Res II 140:151–162

Kaschner K, Tittensor DP, Ready J, Gerrodette T, Worm B (2011) Current and future patterns of global marine mammal biodiversity. PLoS One 6:e19653

Kawecki TJ, Ebert D (2004) Conceptual issues in local adaptation. Ecol Lett 7:1225–1241

Konovalov DA, Manning C, Henshaw MT (2004) KINGROUP: a program for pedigree relationship reconstruction and kin group assignments using genetic markers. Mol Ecol Notes 4: 779–782

Leatherwood S, Jefferson TA, Norris JC, Stevens WE, Hansen LJ, Mullin KD (1993) Occurrence and sounds of Fraser's dolphins (*Lagenodelphis hosei*) in the Gulf of Mexico. Tex J Sci 45:349–354

Leigh JW, Bryant D (2015) PopART: Full-feature software for haplotype network construction. Methods Ecol Evol 6:1110–1116

Librado P, Rozas J (2009) DnaSP v5: A software for comprehensive analysis of DNA polymorphism data. Bioinformatics 25:1451–1452

Louis M, Fontaine MC, Spitz J, Schlund E and others (2014) Ecological opportunities and specializations shaped genetic divergence in a highly mobile marine top predator. Proc R Soc B 281:20141558

MacLeod CD (2009) Global climate change, range changes and potential implications for the conservation of marine cetaceans: a review and synthesis. Endang Species Res 7:125–136

Mayewski PA, Rohling EE, Stager JC, Karlénd W and others (2004) Holocene climate variability. Quat Res 62:243–255

Mignucci-Giannoni AA, Montoya-Ospina RA, Pérez-Zayas JJ, Rodríguez-López MA, Williams EH (1999) New records of Fraser's dolphin (*Lagenodelphis hosei*) for the Caribbean. Aquat Mamm 25: 15-19

Miyazawa Y, Zhang R, Guo X, Tamura H and others (2009) Water mass variability in the western North Pacific detected in a 15-year eddy resolving ocean reanalysis. J Oceanogr 65: 737–756

Moreno IB, Danilewicz D, Borges-Martins M, Ott PH, Caon G, Oliveira LR (2003) Fraser's dolphin (*Lagenodelphis hosei* Fraser, 1956) in Southern Brazil. LAJAM 2:39–46

Moura AE, Nielsen SC, Vilstrup JT, Moreno-Mayar JV and others (2013) Recent diversification of a marine genus (*Tursiops* spp.) tracks habitat preference and environmental change. Syst Biol 62:865–877

Moura AE, van Rensburg CJ, Pilot M, Tehrani A and others (2014) Killer whale nuclear genome and mtDNA reveal widespread population bottleneck during the last glacial maximum. Mol Biol Evol 31:1121–1131

National Museum of Nature and Science (2018) Marine Mammal Stranding Database. www.kahaku.go.jp/english/research/db/zoology/marmam/drift/ (accessed 30 April 2019)

Nei M (1987) Molecular evolutionary genetics. Columbia University Press, New York

Palsbøll PJ, Bérubé M, Allendorf FW (2007) Identification of management units using population genetic data. Trends Ecol Evol 1:11–16

Paradis, E (2010) pegas: an R package for population genetics with an integrated–modular approach. Bioinformatics 26:419–420

Pastene LA, Goto M, Kanda N, Zerbini AN and others (2007) Radiation and speciation of pelagic organisms during periods of global warming: the case of the common minke whale, *Balaenoptera acutorostrata*. Mol Ecol 16:1481–1495

Perrin WF, Best PB, Dawbin WH, Balcomb KC, R. Gambell R, Ross GJB (1973) Rediscovery of Fraser's dolphin *Lagenodelphis hosei*. Nature 241:345–350

Perrin WF, Dolar MLL, Amano M, Hayano A (2003) Cranial sexual dimorphism and geographic variation in Fraser's dolphin, *Lagenodelphis hosei*. Mar Mam Sci 19:484–501

Perrin WF, Reeves RR, Dolar MLL, Jefferson TA, Marsh H, Wang JY, Estacion J (2005) Report of the second workshop on the biology and conservation of small cetaceans and dugongs of South-East Asia. UNEP/CMS Secretariat, Bonn

Porter L, Lai HY (2017) Marine mammals in asian societies; trends in consumption, bait, and traditional use. Front Mar Sci 4:47

Prada C, Schizas NV, Yoshioka PM (2008) Phenotypic plasticity or speciation? A case from a clonal marine organism. BMC Evol Biol 8:47

Renema W, Bellwood DR, Braga JC, Bromfield K and others (2008) Hopping hotspots: global shifts in marine biodiversity. Science 321:654–657

Rinaldi C, Rinaldi R (2011) Les cetaces dans l'archipel guadeloupeen et dans la Caraïbe. Etat des lieux des connaissances et perspectives 1998–2009. Rapport de l'association Evasion Tropicale pour l'Agence des Aires Marines Protégées. Agence des aires marines protégées-Sanctuaire Agoa, Saint Claude

Rogers AR (1995) Genetic evidence for a Pleistocene population explosion. Evolution 49:608–615

Sambrook J, Fritsch EF, Maniatis T (1989) Molecular cloning: a laboratory manual. Cold Spring Harbor Press, Cold Spring Harbor, NY

Schneider S, Excoffier L (1999) Estimation of past demographic parameters from the distribution of pairwise differences when the mutation rates vary among sites:

application to human mitochondrial DNA. Genetics 152:1079–1089

Shen KN, Jamandre BW, Hsu CC, Tzeng WN, Durand JD (2011) Plio-Pleistocene sea level and temperature fluctuations in the northwestern Pacific promoted speciation in the globally-distributed flathead mullet *Mugil cephalus*. BMC Evol Biol 11:83–83

Tseng Y-P, Huang Y-C, Kyle GT, Yang M-C (2011) Modeling the impacts of cetacean-focused tourism in Taiwan: observations from cetacean watching boats: 2002–2005. Environ Manage 47:56–66

Taguchi M, Chivers SJ, Rosel OE, Matsuishi T, Abe S (2010) Mitochondrial DNA phylogeography of the harbour porpoise *Phocoena phocoena* in the North Pacific. Mar Biol 157:1489–1498

Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S (2011) MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Mol Biol Evol 28:2731–2739

Taylor BL, Chivers SJ, Larese J, Perrin WF (2007) Generation length and percent mature estimates for IUCN assessments of cetaceans. Administrative Report LJ-07–01. NOAA Southwest Fisheries Science Center, La Jolla, CA

Tobayama T, Nishiwaki M, Yang HC (1973) Records of the Fraser's Sarawak dolphin (*Lagenodelphis hosei*) in the western North Pacific. Sci Rep Whales Res Inst 25:251–263

Van Bree PJH, Collet A, Desportes G, Hussenot E, Raga JA (1986) Le dauphin de Fraser, *Lagenodelphis hosei* (Cetacea, Odontoceti), espece nouvelle pour la faune d'Europe. Mammalia 50:57–86

Van Oosterhout C, Hutchinson WF, Wills DP, Shipley P (2004) MICRO - CHECKER: software for identifying and correcting genotyping errors in microsatellite data. Mol Ecol Notes 4:535–538

Voris HK (2000) Maps of Pleistocene sea levels in Southeast Asia: shorelines, river systems and time durations. J Biogeogr 27:1153–1167

Wang P (1999) Response of Western Pacific marginal seas to glacial cycles: paleoceanographic and sedimentological features. Mar Geol 156:5–39

Watkins WA, Daher MA, Fristrup K, Notarbartolo-di-Sciara G (1994) Fishing and acoustic behavior of Fraser's dolphin (*Lagenodelphis hosei*) near Dominica, Southeast Caribbean. Caribb J Sci 30:76–82

Weir CR, Debrah J, Ofori-Danson PK, Pierpoint C, Van Waerebeek K (2008) Records of Fraser's dolphin *Lagenodelphis hosei* Fraser 1956 from the Gulf of Guinea and Angola. Afr J Mar Sci 30:241–246

West-Eberhard MJ (1989) Phenotypic plasticity and the origins of diversity. Annu Rev Ecol Syst 20:249–278

Yang S-C, Liao H-C, Pan C-L, Wang JY (1999) A survey of cetaceans in the waters of central-eastern Taiwan. Asian Mar Biol 16:23–34

Yao C-J, Chou L-S, Yang Y-J (2004) Population genetic structure of pantropical spotted dolphin, *Stenella attenuata*, in waters of Taiwan and South China Sea based on mitochondrial DNA control region sequences. Taiwania 49: 80–94

Figure Captions:

Figure 1: Map of the sampling locations. Solid triangles indicate sampling locations, and the numbers in the parentheses indicate the sample size using in microsatellite/mitochondrial DNA analyses.

Figure 2: Result from the discriminant analysis of principal components (DAPC). Individuals represented as dots and the groups as ellipses. For the inset of discriminant analysis (DA) eigenvalues, the x axis represents linear discriminants and the y axis represents the corresponding F-statistics; for the inset of principal component analysis (PCA) eigenvalues, the x axis represents the number of retained PCs and the y axis shows corresponding cumulative variance. CNP = Central North Pacific; ETP = Eastern Tropical Pacific.

Figure 3: Result of the Geneland analysis showing the most common pattern of the population membership when K = 4. The four panels show the landscape of the range likelihood of each population: A) Caribbean Sea; B) Taiwan; C) the Philippines, central-eastern tropical Pacific, and Gulf of Mexico; and D) Japan. Note that the population shown in panel C was sporadically distributed in multiple locations. The dots represent the samples, with geographical locality indicated in the top panel. Probability values shown on contour lines and indicated by colours: red as low probability and white as high probability.

Figure 4: Median-joining network plot showing the relationship among the mtDNA control region haplotypes. The circles represent unique haplotypes with different colour shades showing the composition of sample origins, and the size indicative of

the number of individuals with that haplotype (see key). Solid black circles indicate missing intermediate haplotypes, and the hatch marks at the lines indicate the number of mutational steps separating the haplotypes.

Table 1: Genetic variability of the 18 microsatellite loci examined in our samples.

Geographic group	n	Missing data rate	No. of alleles	Ave. H_E	Ave. H_O	Ave. F
Japan	37	0.15%	115	0.6	0.61	0.165
Taiwan	43	0.78%	137	0.62	0.54	0.214
Philippines	17	1.31%	92	0.57	0.62	0.147
Central North Pacific (CNP)	3	0%	52	0.59	0.59	
Gulf of Mexico (GM)	2	0%	42	0.57	0.58	
Caribbean Sea (CS)	3	0%	39	0.43	0.46	
Eastern Tropical Pacific (ETP)	1	0%	30	NA	NA	
All samples	106	0.58%		0.61	0.58	

Table 2: Pairwise genetic differences among the three main groups according to microsatellite data: above diagonal, F_{ST}; below diagonal, 95% confidential interval.

	Japan	Taiwan	Philippines
Japan		0.0085	0.0133
Taiwan	0.003—0.015		0.0103
Philippines	0.005—0.021	-0.002—0.025	

Table 3: Haplotype counts, genetic diversity, nucleotide diversity, Tajima's D and Fu's Fs estimates of a 779bp mtDNA control region sequence in the samples. All sequences include samples from Central North Pacific, Eastern Tropical Pacific, Gulf of Mexico and Caribbean Sea.

Geographic group	n	Number of variable sites	Number of haplotypes	Haplotype diversity (h)	Nucleotide diversity (π)	Average number of nucleotide differences (k)	Tajima's D	Fu's Fs
Japan	35	44	24	0.973 (0.014)	0.012 (0.10%)	9.689	-0.41	-6.834**
Taiwan	42	40	22	0.958 (0.013)	0.012 (0.07%)	9.417	-0.041	-3.197*
Philippines	10	26	7	0.911 (0.077)	0.012 (0.21%)	9.044	-0.076	0.64
Western North Pacific	87	61	42	0.973 (0.006)	0.012 (0.06%)	9.534	-0.777	-14.233***
All sequences	96	64	46	0.974 (0.005)	0.012 (0.05%)	9.588	-0.824	-17.243***

***: $p < 0.001$, **: $p < 0.01$, *: $p < 0.05$.

Table 4: Mismatch analysis results for A) demographic expansion and B) spatial expansion models. τ is the time since expansion measured in mutational time units, SSD is the sum of squared deviation in goodness-of-fit test, and r is the raggedness index. T_1 and T_2 are the time of demographic/spatial changes for each geographic group calculated using substitution rates (μ) of $1x10^{-7}$ and $7x10^{-8}$, respectively. The 95% profile likelihood for the estimates is given in parentheses.

Geographic group	τ (95% CI)	SSD	r	T_1 (95% CI)	T_2 (95% CI)
A) Demographic expansion model					
Japan	13.4 (7.254—17.988)	0.012	0.014	7748 (4195—10401)	11069 (5992—14859)
Philippines	12.6 (4.996—17.707)	0.023	0.044	7286 (2889—10239)	10408 (4127—14627)
Taiwan	11.5 (5.68—19.568)	0.005	0.011	6650 (3284—11315)	9500 (4692—16164)
Western North Pacific	13.1 (6.051—18.041)	0.003	0.004	7575 (3499—10432)	10821 (4998—14903)
B) Spatial expansion model					
Japan	8.396 (4.8—20.161)	0.021	0.014	4855 (2776—11658)	6936 (3965—16654)
Philippines	9.042 (5.105—18.239)	0.026	0.044	5228 (2952—10547)	7469 (4217—15067)
Taiwan	7.551 (4.547—19.242)	0.01	0.011	4366 (2629—11127)	6238 (3756—15895)
Western North Pacific	7.091 (4.265—20.619)	0.009	0.004	4100 (2466—11923)	5858 (3523—17033)

Table 5: Pairwise divergence between the three main geographic groups according to mtDNA data.

		F_{ST}		
		Japan	Taiwan	Philippines
Φ_{ST}	Japan		0.01	0.029*
	Taiwan	0.009		0.034*
	Philippines	0.031	-0.017	

*: $p < 0.05$

Figure 1:

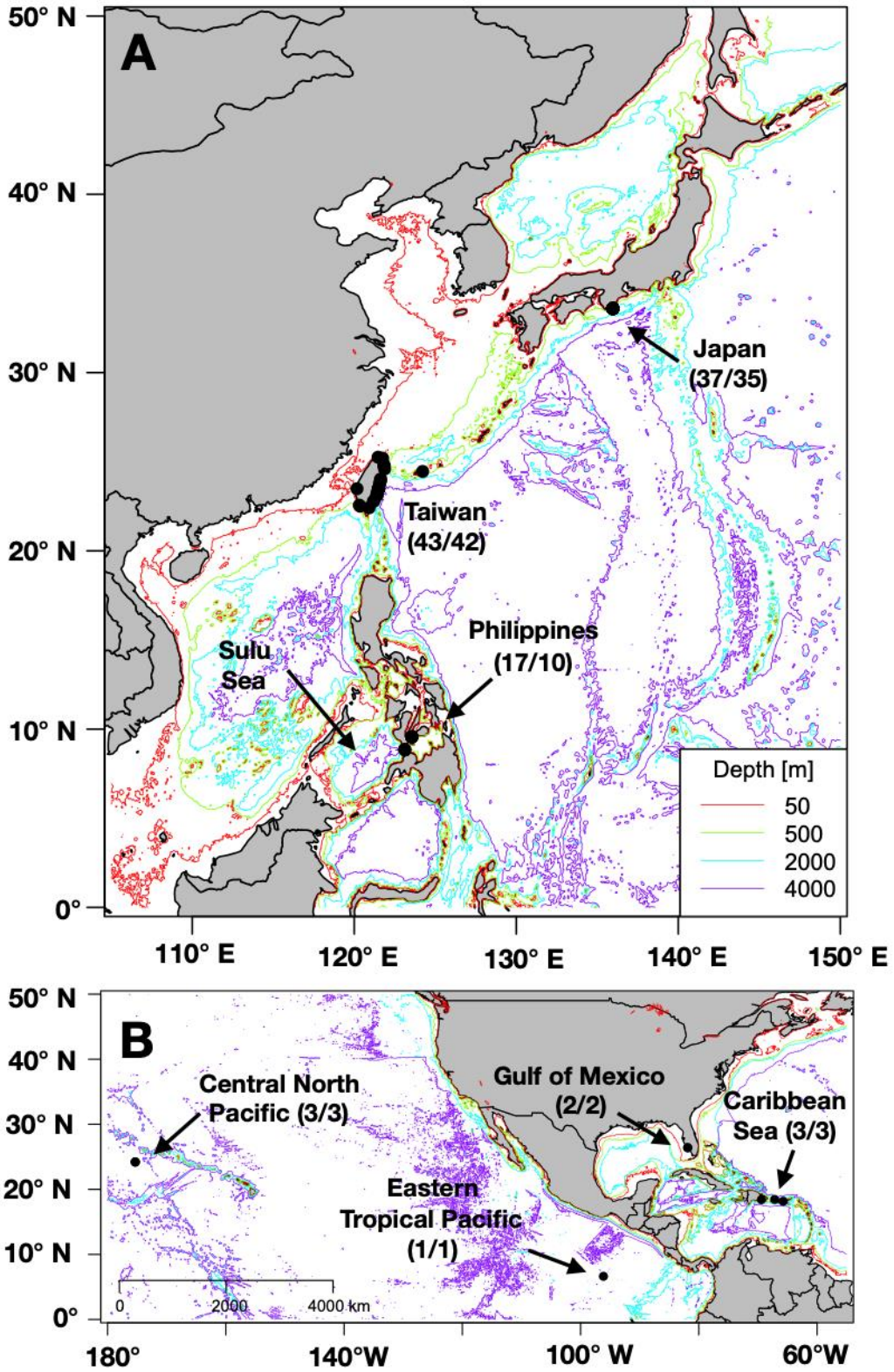
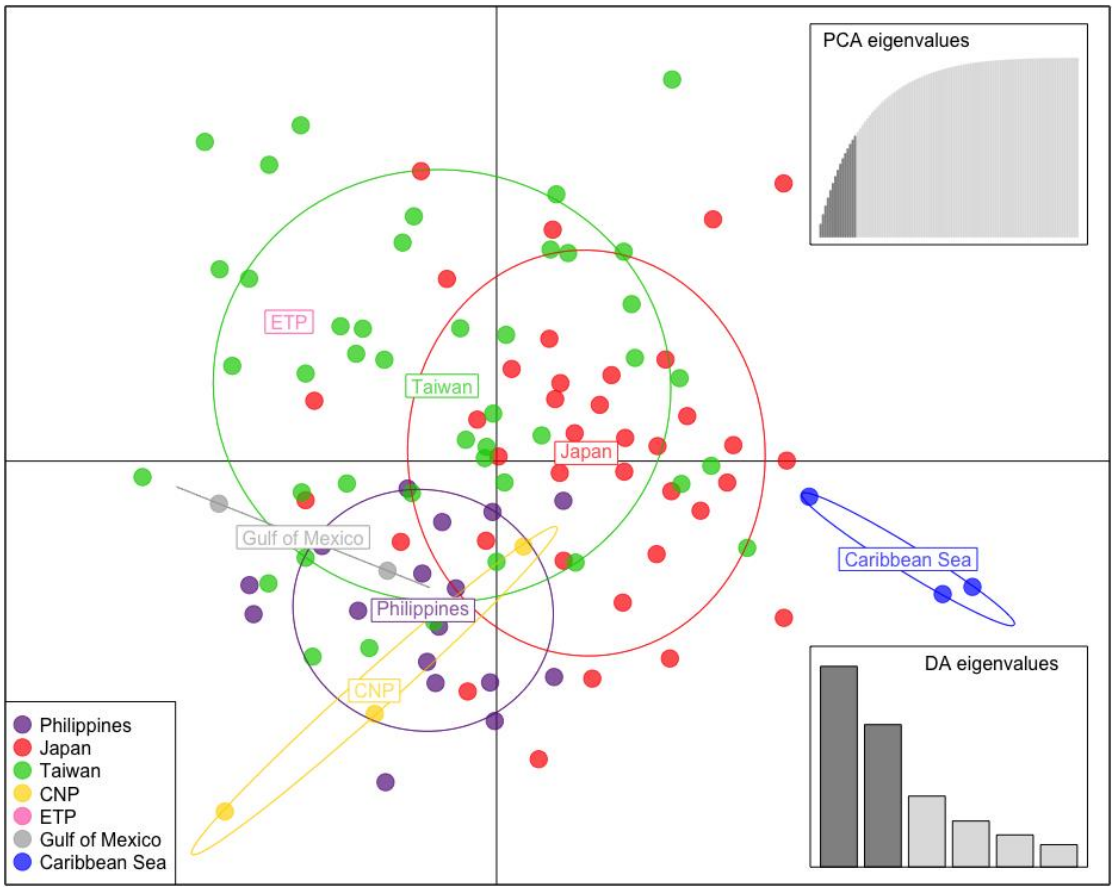

A
50° N
40° N
30° N
20° N
10° N
0°
110° E
120° E
130° E
140° E
150° E
Japan
(37/35)
Taiwan
(43/42)
Philippines
(17/10)
Sulu
Sea
Depth [m]
50
500
2000
4000
B
50° N
40° N
30° N
20° N
10° N
0°
180°
140°W
100° W
60°W
Central North
Pacific (3/3)
Gulf of Mexico
(2/2)
Caribbean
Sea (3/3)
Eastern
Tropical Pacific
(1/1)
0
2000
4000 km

Figure 2:


PCA eigenvalues
ETP
Taiwan
Japan
Gulf of Mexico
Caribbean Sea
Philippines
CNP
DA eigenvalues
Philippines
Japan
Taiwan
CNP
ETP
Gulf of Mexico
Caribbean Sea

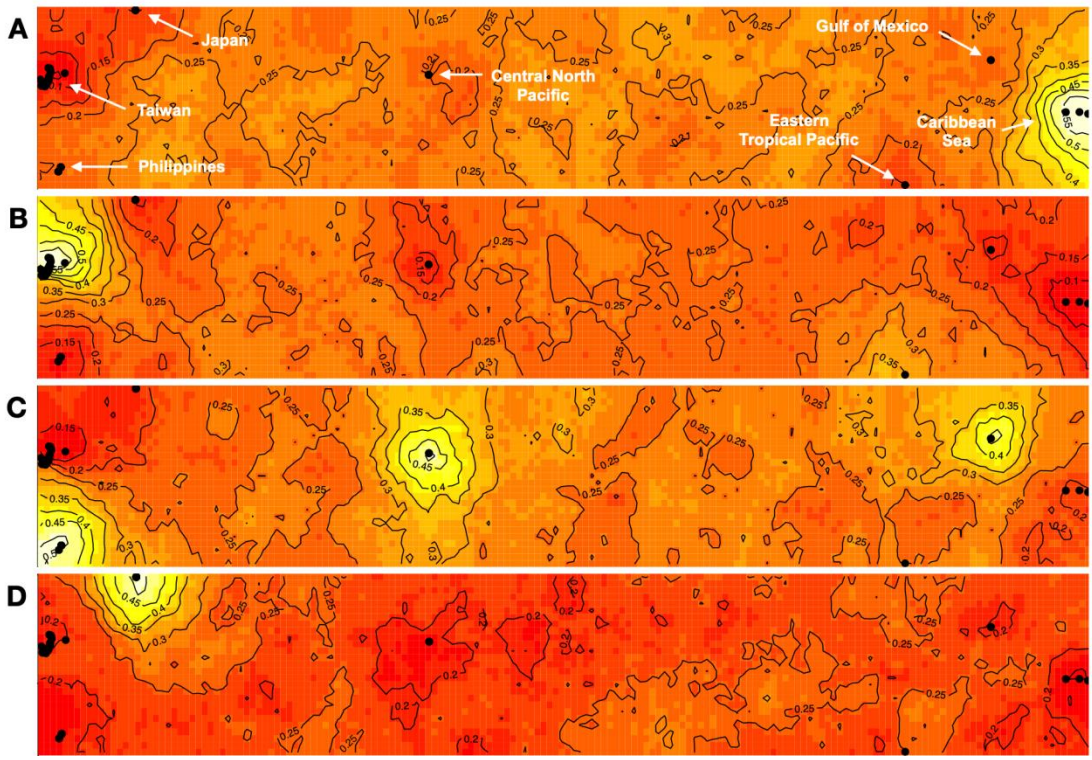
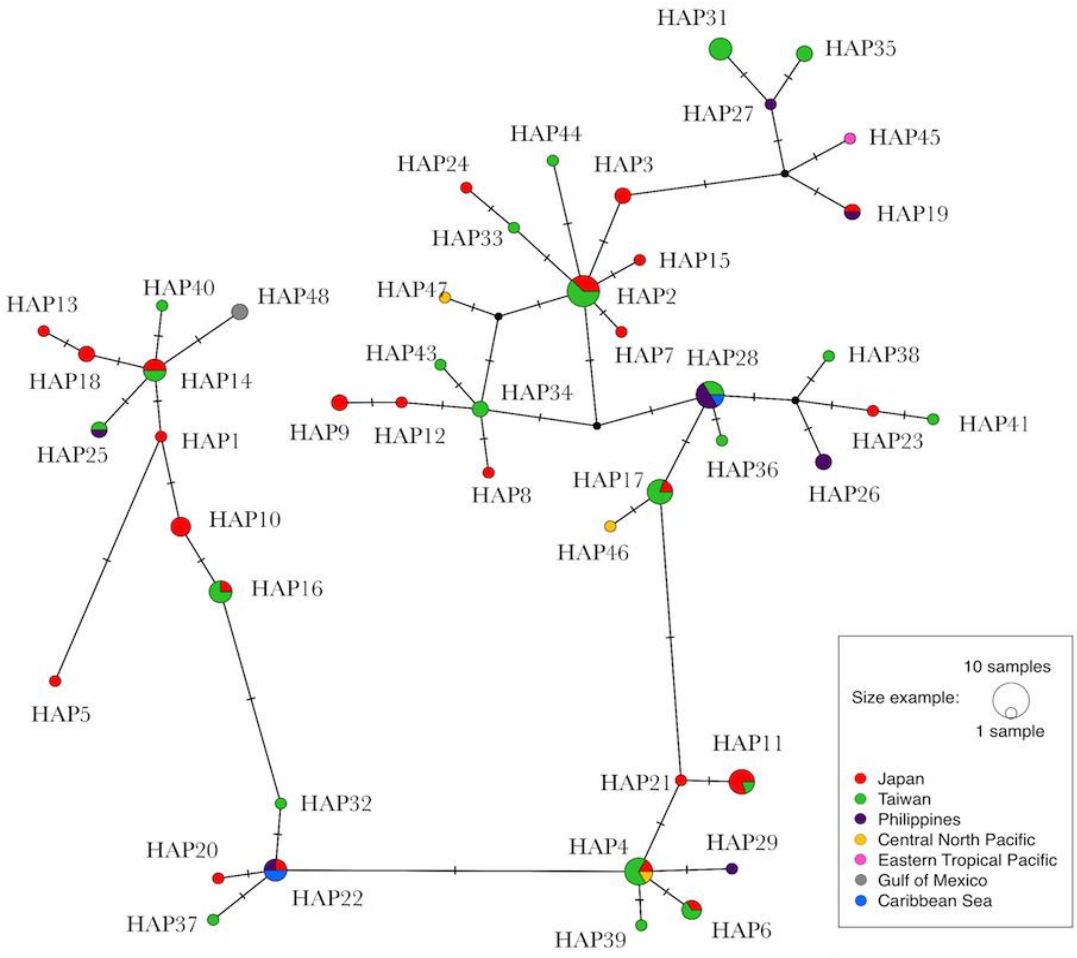

A
B
C
D
Japan
Taiwan
Philippines
Central North Pacific
Gulf of Mexico
Eastern Tropical Pacific
Caribbean Sea

Figure 3:

Figure 4:


HAP31
HAP35
HAP27
HAP45
HAP44
HAP24
HAP3
HAP19
HAP33
HAP15
HAP40
HAP48
HAP13
HAP47
HAP2
HAP7
HAP43
HAP28
HAP38
HAP18
HAP14
HAP34
HAP41
HAP9
HAP12
HAP23
HAP25
HAP1
HAP36
HAP8
HAP17
HAP26
HAP10
HAP46
HAP16
HAP5
HAP11
HAP21
HAP32
HAP20
HAP4
HAP29
HAP22
HAP37
HAP39
HAP6
10 samples
Size example:
1 sample
Japan
Taiwan
Philippines
Central North Pacific
Eastern Tropical Pacific
Gulf of Mexico
Caribbean Sea